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High throughput 16S rRNA gene amplicon sequencing

A fast and cheap method to study the influence of microbial community composition on activated sludge floc properties

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High throughput 16S rRNA gene amplicon sequencing: a fast and cheap method to study the influence of microbial community composition on activated sludge properties



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Introduction

A reliable and reproducible method for identification and quantification of microorganisms is important for the studies of microbial communities in activated sludge and for the demonstration of their significance for plant operation and stability.

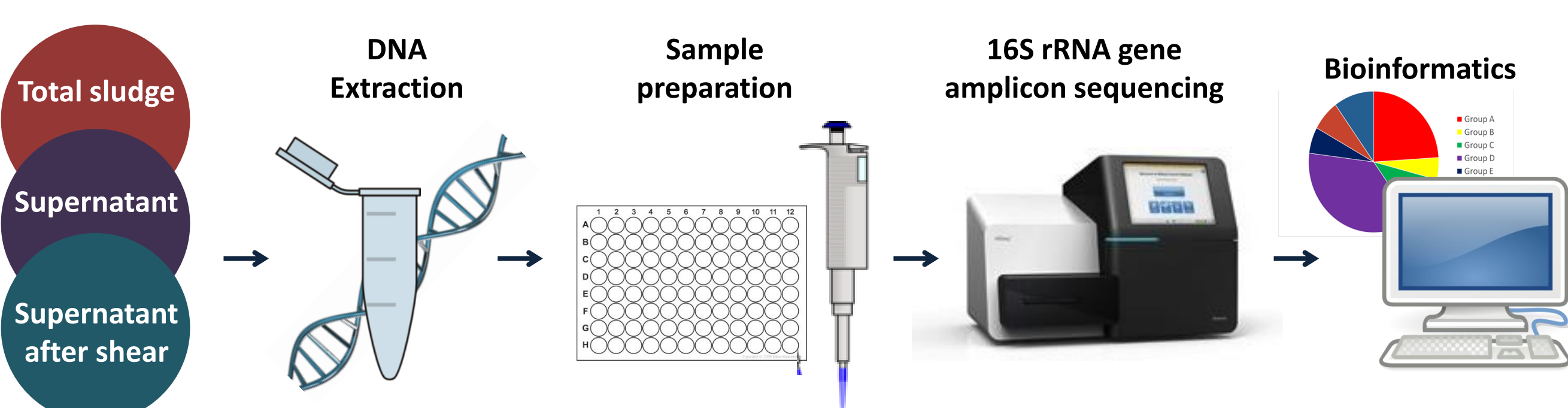
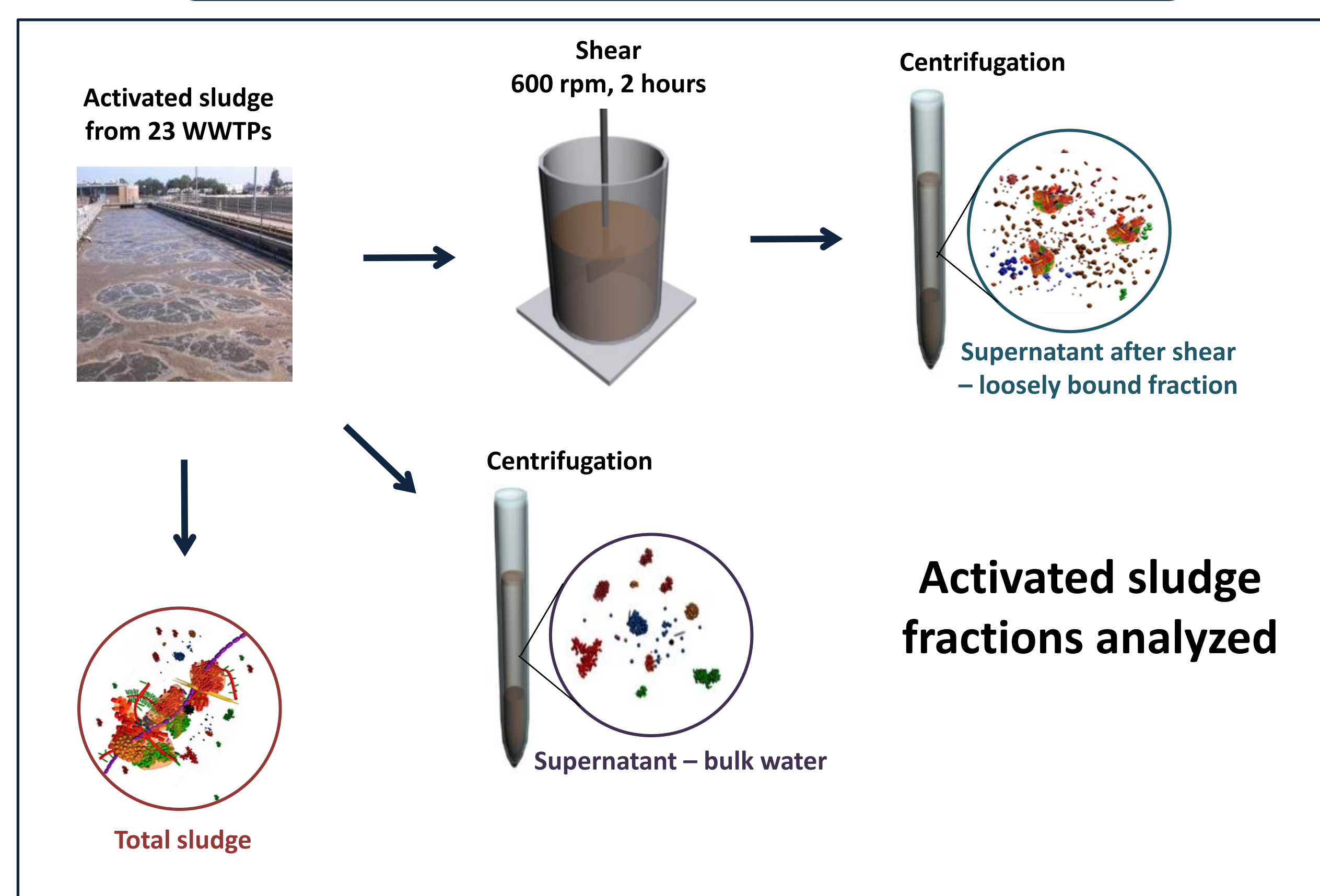
DNA-based identification of microorganisms using 16S rRNA gene amplicon sequencing has been developed over the past few years and is now ready to use for more comprehensive studies related to plant operation and optimization thanks to short analysis time, low cost, high throughput, and high taxonomic resolution.

Since bacterial morphology, mode of growth and EPS composition determine floc size, shape and strength, which in turn influence important sludge properties, the link established between the microbial community structure and physico-chemical sludge characteristics may provide a better understanding of the activated sludge process.

Objectives

- ❑ To show how 16S rRNA gene amplicon sequencing can be used to reveal factors of importance for the operation of 23 full-scale nutrient removal plants that can be related to settling problems and floc properties.
- ❑ To investigate whether the microbial community composition differs between the flocs and the supernatant (bulk water) and whether certain bacterial species are prone to detachment from the flocs.

Methods



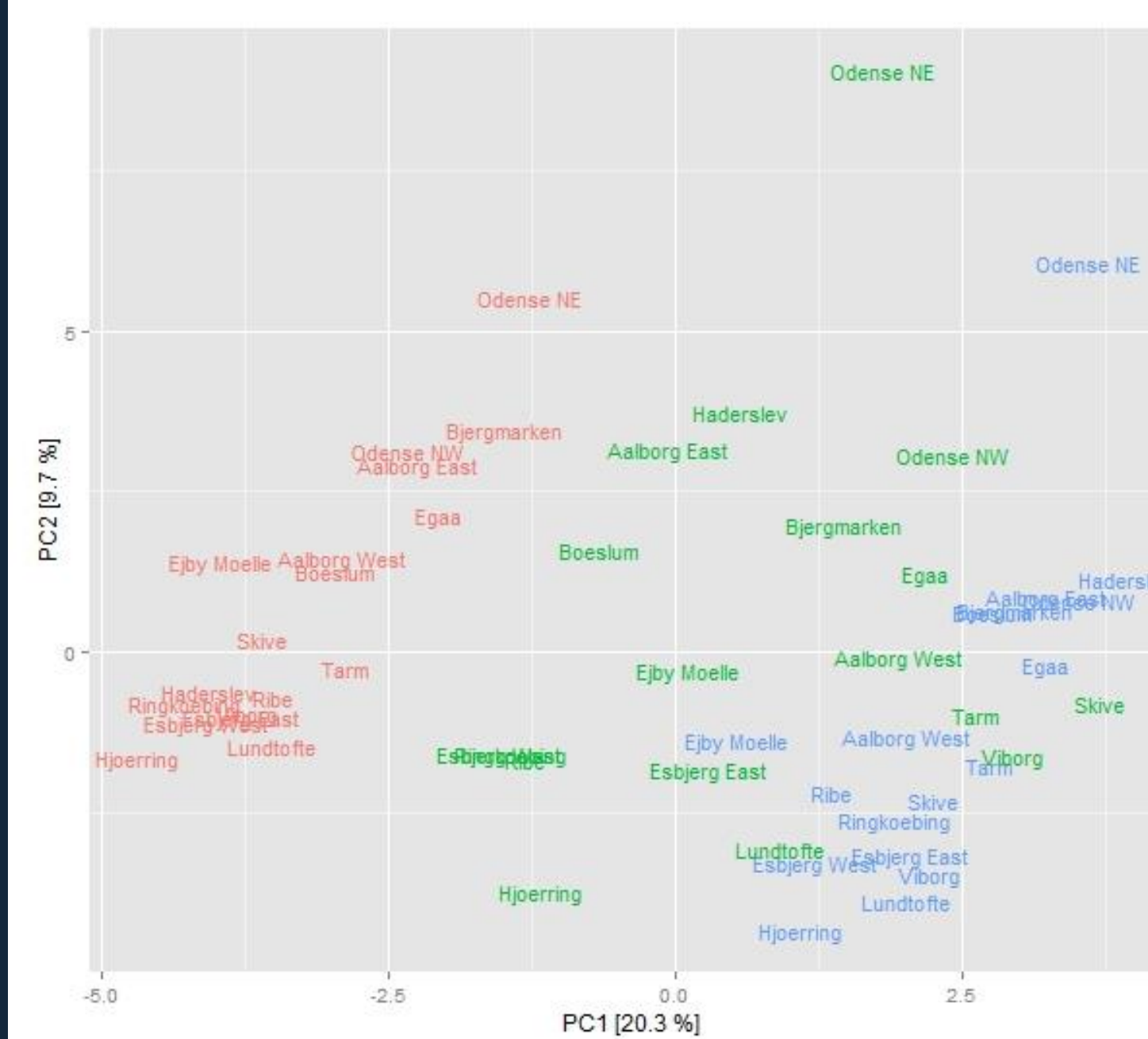
Conclusions

16S rRNA gene amplicon sequencing is suitable for comprehensive studies of WWTPs thanks to short analysis time, low cost, high throughput, and high taxonomic resolution.

A number of bacterial species can be correlated to the sludge characteristics that are important for the proper plant operation (SVI, floc strength, and EPS content).

Specific bacteria are enriched in the bulk water fraction and in the fraction loosely bound to the floc.

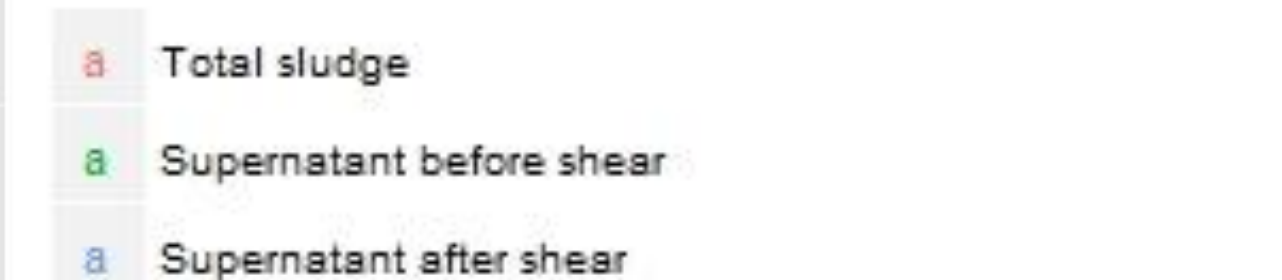
Results



Community composition in different activated sludge fractions

The figure shows the relationship between all samples analyzed from 23 WWTPs.

Samples grouping into individual sludge fractions (total sludge – bulk water – loosely bound fraction) can be clearly observed.

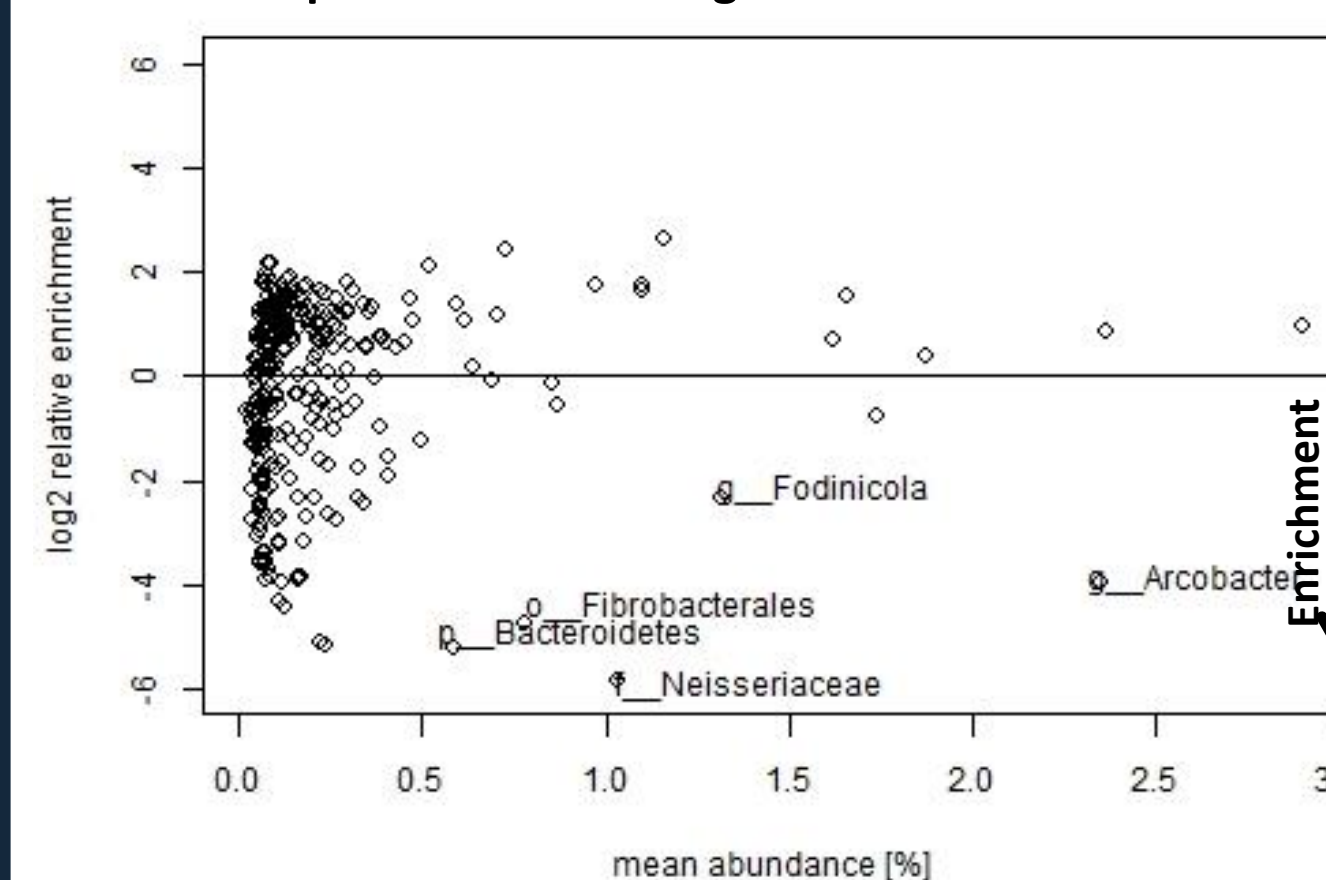


Relative abundance

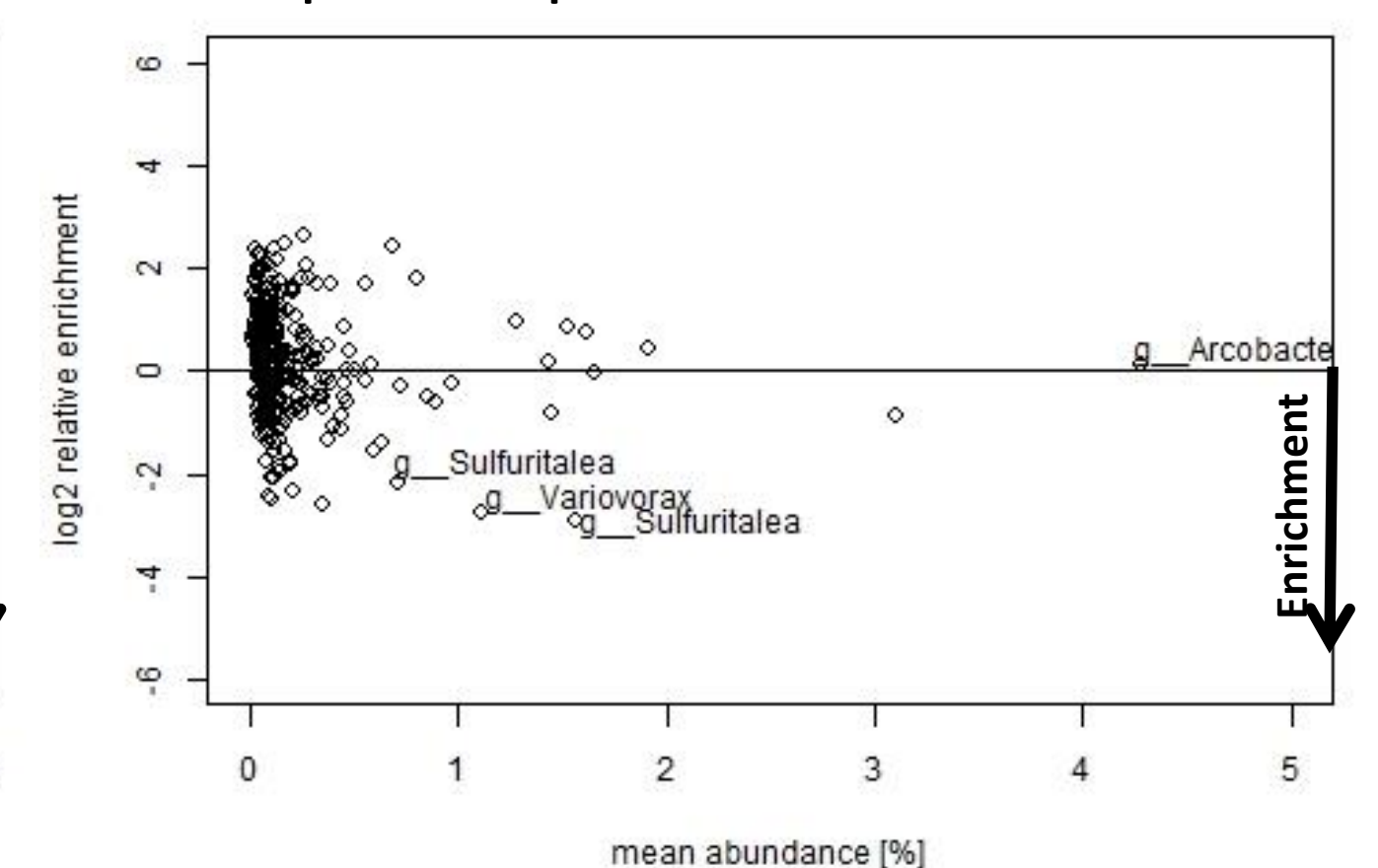
of 10 most frequently occurring genera compared between the different sludge fractions.

The variation between the WWTPs is captured by the boxplot width.

Bacteria enriched in the supernatant compared to total sludge



Bacteria enriched in the supernatant after shear compared to supernatant

[illegible][illegible]

Spearman correlation of bacteria present in different sludge fractions with important sludge characteristics: Sludge Volume Index (SVI), shear sensitivity, degree of flocculation and conditions for flocculation. Bacteria that were highly correlated ($R > 0.6$) with the mentioned parameters are listed above.